

Identification of multipotent stem/progenitor cells in murine sclera.

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Public Summary:

The sclera is a dense fibrous and viscoelastic connective tissue. It forms the outer coat frame of the eye to keep the shape of eyeball by enduring the pressure from outside and inside. In eutherian mammals, the entire sclera consists of fibrous and type I collagen-dominated extracellular matrix. Recent research has showed that the sclera is a dynamic tissue, capable of responding rapidly to changes in the visual environment to alter ocular size and refraction. Sclera's biochemical and biomechanical properties determine the shape and size of the eyeball and therefore play an important role in determination of refraction status. Scleral extracellular matrix remodeling is responsible for changes in ocular elongation disease such as myopia. Diseases in sclera such as refractory scleritis and scleromalacia are all difficult to cure and often lead to blindness. It is speculated that sclera cells might play an important role in scleral remodeling or sclera inflammation. Although tissue-specific postnatal stem cells have been isolated from a variety of organs and tissues, it is still unknown whether sclera contains stem/progenitor cells. The purpose of this study is to identify multipotent scleral stem/progenitor cells (SSPCs) from murine sclera. We found that SSPCs possessed clonogenic and self-renewing capacities. They were positive for mesenchymal markers Sca1, CD90.2, CD44, CD105, CD73, and negative for hematopoietic markers CD45, CD11b, Flk1, CD34, CD117. In addition to express stem cell genes ABCG2, Six2, Notch1 and Pax6, SSPCs were able to differentiate to adipogenic, chondrogenic, and neurogenic lineages. This study indicated that sclera contained multipotent mesenchymal stem cells SSPCs. Further study of SSPCs may benefit understanding molecular mechanism of sclera diseases such as scleritis and myopia.

Scientific Abstract:

PURPOSE: The sclera forms the fibrous outer coat of the eyeball and acts as a supportive framework. The purpose of this study was to examine whether the sclera contains mesenchymal stem/progenitor cells. **METHOD:** Scleral tissue from C57BL6/J mice was separated from the retina and choroid and subsequently enzyme digested to release single cells. Proliferation capacity, self-renewal capacity, and ability for multipotent differentiation were analyzed by BrdU labeling, flow cytometry, reverse transcriptase-polymerase chain reaction, immunocytochemistry, and in vivo transplantation. **RESULTS:** The scleral stem/progenitor cells (SSPCs) possessed clonogenic and high doubling capacities. These cells were positive for the mesenchymal markers Sca-1, CD90.2, CD44, CD105, and CD73 and negative for the hematopoietic markers CD45, CD11b, Flk1, CD34, and CD117. In addition to expressing stem cell genes ABCG2, Six2, Notch1, and Pax6, SSPCs were able to differentiate to adipogenic, chondrogenic, and neurogenic lineages. **CONCLUSIONS:** This study indicates that the sclera contains multipotent mesenchymal stem cells. Further study of SSPCs may help elucidate the cellular and molecular mechanism of scleral diseases such as scleritis and myopia.

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